



Age-Related Distribution of Basal Anti-Mullerian Hormone Levels in a Population of Infertile Women

İnfertil Kadınlarının Bazal Anti-Müllerian Hormon Serum Seviyelerinin Yaşa Özgü Dağılımı

Age Related Distribution of AMH Levels

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Özet

Amaç: Mevcut araştırmada kadınlarda serum Anti-Müllerian Hormon (AMH) ölçümlerinin yaşa özgü referans değerlerini belirlemek, yaşa özgü beklenen değerlerle ilgili bir referans değeri oluşturmak ve çocuk sahibi olmak amacıyla infertilite polikliniğine başvuran doğurgan yaştaki Türk kadınlarının bazal serum AMH seviyelerinin dağılımını analiz etmeyi amaçladık. **Gereç ve Yöntem:** Türkiye'deki tek bir infertilite merkezine 12 aylık periyot içinde başvuran 409 kadının siklusun 2-3. gününde prospektif olarak toplanan AMH ölçümlerinin retrospektif analizi yapılmıştır. **Bulgular:** Serum AMH konsantrasyonu yaşla birlikte azalma göstermiştir. Yaş ortalaması 34.04±5.39, serum AMH konsantrasyon ortalaması 1.77±1.82 idi. AMH seviyeleri 20-24, 25-29, 30-34, 35-39, ve >40 yaş gruplarına göre incelendi. Ortanca AMH değerleri belirtilen yaşlara göre sırası ile 2.16 ng/ml, 2.15 ng/ml, 1.71 ng/ml, 0.80 ng/ml ve 0.47 ng/ml'dir. **Tartışma:** Çalışmada sunulan mevcut veriler Türk infertilite kadınlarda serum AMH konsantrasyonu için bir çerçeve oluşturacaktır.

Anahtar Kelimeler

Anti-Müllerian Hormon; Reprodüktif Yaş; Yaşa Özgü Değerler

Abstract

Aim: We aimed to constitute age-specific reference serum values for anti-Mullerian Hormone (AMH) in women, and to analyze the distribution of basal serum AMH levels in Turkish women of reproductive age attending an infertility clinic to provide a framework for expected values according to age. **Material and Method:** Retrospective analysis of prospectively collected data on cycle day 2-3 serum AMH measurements of 409 women attending a single infertility unit in Turkey through a 12-month-period was performed. **Results:** Concentrations of serum AMH were shown to decrease with advancing age of the female partner. The mean age of the women was 34.04±5.39 years and the mean AMH level of the women was 1.77±1.82. The AMH levels were grouped according to age as follows: 20-24, 25-29, 30-34, 35-39, and >40 years. The median AMH values were 2.16 ng/ml, 2.15 ng/ml, 1.71 ng/ml, 0.80 ng/ml, and 0.47 ng/ml, respectively according to the age groups. **Discussion:** The present data provide a framework for age-specific serum AMH levels in a Turkish population of infertile women.

Keywords

Anti-Mullerian Hormone; Reproductive Age; Age-Specific Values

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Introduction

The dimeric glycoprotein AMH, a member of the transforming growth factor- β (TGF- β) superfamily, is produced by granulosa cells especially during the preantral and small antral follicle stages, independent of follicle stimulating hormone (FSH) [1]. Recent studies demonstrated that AMH is one of the most reliable predictor of the ovarian reserve and ovarian response to stimulation in assisted reproductive technologies (ART) [2-5]. AMH is also recognized as a good indicator for potential fertility. Therefore, AMH can facilitate counseling of patients and consideration of treatment options for individualized management and set expectations of practitioners in ART. The serum AMH concentration is inversely correlated with increasing age, and thus serum concentrations of AMH decrease with advanced age accompanied by a concomitant decline in the number of primordial follicles [6,7]. Natural fertility starts to decline after the age of 30, but it is very likely that the process of follicle decline varies individually [8]. Serum AMH levels could be routinely measured during clinical fertility evaluations and utilized in counseling women who wished to postpone childbearing. We are not aware of an age-specific median of AMH among a population of infertile Turkish women that could be used as a reference value. Therefore, the aim of the current study was to calculate age-specific serum AMH levels and analyze the distribution of basal serum AMH levels in infertile Turkish women of reproductive age attending an infertility clinic to provide a framework for expected age-related values..

Material and Method

Participants

This retrospective study was based on the analysis of medical records. Of 580 women admitted to an IVF unit with the complaint of infertility between April 2012 and March 2013, 409 women who met the inclusion criteria were recruited for the study. The study was conducted at an IVF center in ** Hospital, Istanbul, Turkey. Written informed consent was obtained from all the study participants. The inclusion criteria were (1) Body-mass index of 19-30 kg/m², (2) age of 20-44 years, (3) primary infertility, (4) both ovaries present on a transvaginal ultrasound scan, (5) no previous history of ovarian surgery, (6) no exposure to cytotoxic drugs or pelvic radiation, and (7) no hormonal therapy in the preceding 6 months. Each woman was represented only once in the study. Women with polycystic ovary syndrome were excluded from this study. Data were collected on age and AMH levels. The AMH levels were grouped according to age as follows: 20-24, 25-29, 30-34, 35-39, and >40 years.

AMH measurements

Serum AMH levels obtained from venous blood by routine methods were measured on day 2 or 3 of a spontaneous cycle within 3 months of commencing ovarian stimulations. AMH levels were determined at a single reference laboratory with an ultrasensitive ELISA (Beckman-Coulter, Villepinte, France) as described elsewhere [9]. The basal serum AMH values are presented in a concentration of ng/ml. The assay range for AMH was 0.16 - 20 ng/mL, functional sensitivity was 0.08 ng/mL; and the intra-assay and interassay coefficients of variation were 5.4% and 5.6%, respectively.

Statistical Analyses

Analyses were done using the Statistical Package for the Social Sciences, version 21 (SPSS, Chicago, IL, USA) and MedCalc version 10.2. Data were expressed as the mean \pm standard deviations, median or frequencies and percentages. Normally distributed parametric variables were tested with Kolmogorov-Smirnov test and Shapiro-Wilks test. The correlation between two different normally distributed parameters was calculated using Pearson's correlation coefficient. $P < 0.05$ was considered significant.

Results

The mean age of the women was 34.04 ± 5.39 years and the mean AMH level of the women was 1.77 ± 1.82 . The median AMH values were 2.16 ng/ml (n=18) in the 20-24 years' age group, 2.15 ng/ml (n=70) in the 25-29 years' age group, 1.71 ng/ml (n=135) in the 30-34 years' age group, 0.8 ng/ml (n=130) in the 35-39 years' age group, and 0.47 ng/ml (n=56) in the over 40 years' age group (Table 1). The distribution of the AMH values among the women according to age is shown in Figure 1. The median AMH levels decreased steadily in a manner highly correlated with advancing age ($p < 0.001$). Of all the women, 41.8% (n=171) had a serum AMH level of ≤ 1 ng/ml. Of those, 39.2% (n=67) were ≤ 35 years (Table 2).

Table 1. Median serum AMH levels of according to different age groups.

Ages	Median serum AMH level (ng/ml)
20-24	2,16
25-29	2,15
30-34	1,71
35-39	0,8
≥ 40	0,47

AMH: Anti-mullerian hormone

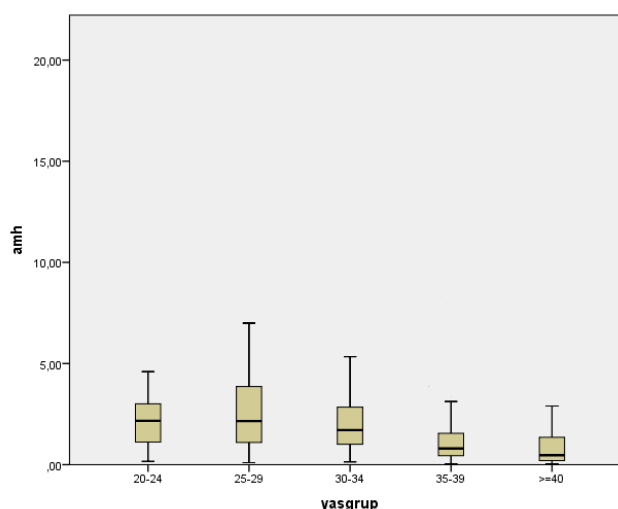


Figure 1. The distribution of AMH values among women of different age groups

Table 2: Serum AMH levels according to cutoff level of 35 years-old

Serum AMH level (ng/ml)	≤ 35 years-old n=247 (60,4%)	> 35 years-old n= 162 (39,6%)
≤ 1	67 (39,2%)	104 (60,8%)
> 1	180 (75,6%)	58 (24,4%)

Discussion

The introduction of AMH in reproductive endocrinology can be considered a revolution in the prediction of the ovarian reserve and response, resulting in the individualization of treatment strategies and counseling of couples regarding the likelihood of the success of IVF treatment [10,11]. There is a trend toward using AMH to individualize treatment strategies and optimize the overall efficacy of IVF treatment. AMH is produced by the granulosa cells especially during preantral and small antral follicle stages. Therefore, serum AMH levels gradually decrease with advancing age due to reduction of the number of antral follicles and it is feasible that AMH can directly assess the follicle pool [12,13]. On the otherhand, endocrinological markers are used to indirectly measure the follicle pool such as FSH. FSH release by the pituitary gland indirectly reflect the number of antral follicles because of the negative feedback action of inhibins, E2, and other ovarian FSH-modulating proteins [14]. Although there are other tests available to predict ovarian reserves, including the antral follicle count and basal FSH levels, serum AMH measurements have many characteristics, such as less dependence upon operators and less intra- and intercycle variation, that make them favorable for use in the clinical setting. An added advantage of AMH measurements is that the blood can be stored, and the serum concentration of AMH can be analyzed at a later stage in stored blood [15-17].

The major drawback with AMH measurements used as an optimal indicator of potential fertility is the obvious lack of reference values for different demographic and ethnic groups. The main aim of our study was to provide age-specific serum AMH reference values and analyze the distribution of basal serum AMH levels in women of reproductive age. In a retrospective study, Shebl et al. [18] evaluated the age-related distribution of basal serum AMH levels in women of reproductive age and concluded that there was wide variation in serum AMH levels in each year of age analyzed, with even young women at risk of reduced ovarian reserves. To the best our knowledge, this is one of the first studies to date to examine age-specific medians, for serum AMH in women undergoing evaluations at Turkish fertility centers.

In the largest study analyzing age-specific medians for serum AMH, Seifer et al [19] reported that both median and mean AMH values were inversely associated with age, and that the average yearly decrease in the median serum AMH value was 0.2 ng/ml/year through age 35 before diminishing to 0.1 ng/mL/year after the age of 35. The rate of decline in mean AMH values was 0.2 ng/ml/year through age 40 and then diminished to 0.1 ng/ml/year thereafter. In their study, the rate of decline in mean AMH values was 0.2 ng/ml/year through age 40, with AMH values diminishing to 0.1 ng/ml/year thereafter. In a longitudinal observational study, de Vet et al. [11] investigated whether serum concentrations of AMH had the potential to be used as a marker for ovarian aging. In the interval between two visits, which ranged from 1.1 to 7.3 years (mean, 2.6 ± 1.7 years), the concentrations of AMH decreased significantly over time (median value, 2.1 g/l at visit 1 vs. 1.3 g/l at visit 2), whereas the number of antral follicles and levels of FSH and inhibin B did not change. They concluded that serum concentrations of AMH decreased over time in young normo-ovulatory

women, whereas other markers associated with ovarian aging did not change.

Our results revealed that the AMH concentration declined significantly with increasing age. This decline began at the age of 30, and it became dramatically evident from the age of 35. This suggests that some women may be candidates of poor response due to the unexpected risk of a diminishing ovarian reserve after age 30. Furthermore, one of parameters of first realistic definitions for standardization of a poor response includes a serum AMH level of 0.5–1.1 ng/ml [21]. Interestingly, almost 40% of women with AMH ≤1 ng/ml (a strong criterion for a poor response to IVF treatment) in our population, were ≤35 years.

Some limitations of the present study need to be mentioned. Our population included only women admitted to an IVF unit with the complaint of infertility, and not healthy women. Therefore, ours was not a community-based study with proper sampling that included healthy, subfertile, and infertile women. This obviously led to a sampling bias. Another drawback was the relatively small sample size. Despite these limitations, our results could provide a reference guide for clinicians in Turkey who counsel women about the timing to become pregnant.

Conclusion

Our data are consistent with the literature, and they provide further evidence for understanding the age-dependent distribution of serum AMH levels in women of reproductive age in Turkey. They might make the serum AMH level particularly attractive for clinical applications and a novel marker for ovarian aging. Further trials with larger numbers of healthy and infertile women are needed to verify these results.

Competing interests

The authors declare that they have no competing interests.

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